(2) The morphology of *Azot. indicum* and *Azot. lacticogenes* is completely different from other species of *Azotobacter*. *Azot. indicum* is a motile rod with usually two fatty bodies at each end of the cell. *Azot. lacticogenes* is a non-motile coccobacillus containing fatty bodies. Physiologically they can grow in acid conditions at a pH as low as 3·5, and produce acid in their media. They do not produce any cyst. *Azot. indicum* produces a slimy substance but *Azot. lacticogenes* does not. The pigments produced by both are different from those of other species of *Azotobacter*. *Azot. lacticogenes* requires a very narrow temperature range (20–30°C.).

It is clear that there is no reason to classify rod or cocci-shaped organisms into the genus *Azotobacter*, which has a yeast-like morphology.

Derx already referred to a new genus, *Beijerinckia*. Later he successfully isolated a new species, *Beij. mobile*, and a new variety, *Beij. indica* var. *alba* (1951).

Azot. lacticogenes is also a *Beijerinckia* which has been mistaken for an *Azotobacter*.* The species becomes *Beij. lacticogenes* (Kauffmann and Toussaint) Tchan, comb. nov., syn. *Azot. lacticogenes*.

Beijerinckia provisionally could be included in the family Azotobacteriaceae. They are relatively large Gram-negative organisms. At least, in certain stages of their development their morphology shows somewhat oval shape cells and evolution forms. Further information is needed to clarify this point.

After this critical examination of morphology and physiology of the representatives of the genus *Azotobacter* the following classification is proposed.

AZOTOBACTERIACEAE Bergey, Bree, Murray, 1938.

Cells relatively large rods or cocci, sometimes almost yeast-like in appearance, especially in media with sugar, motile or non-motile, Gram-negative.

The family has three genera:

1. *Azotobacter* Beijerinck, 1901.

Cells rod or oval shaped, motile or non-motile; when motile, ciliation is peritrichous. In media with alcohol or organic acid as the energy source, cysts are formed. Acid is usually not produced in the media containing sugar. N is fixed in media free of combined N. Aerobic.

Three species and one variety are accepted.

Species typica, *Azotobacter chroococcum* Beijerinck.

Distinguishing Characters for Different Species.

	Morphology.	Motility.	Pigmentation.	Physiology.	Habitat.
Azot. chroococcum	Oval or rod. 2–3×1·5 μ	+	Brown black.	Growth inhibited by 1% of benzoate but uses benzoate at 0·5%.	Soil.
Azot. vinelandii	Rod. 2·5×1·5 μ	+	Green fluorescent.	Uses benzoate at 1% or less.	Soil, water.
Azot. beijerinckii	Oval. 3·3–25×2 μ	–	Yellow.	Uses benzoate at 0·5% but growth inhibited by 1% benzoate.	Soil.
Azot. bei. var *acido-tolerans*	,,	,,	,,	*Id.* except the tolerance to acidity of media.	Soil.

* Dr. Kauffmann has kindly provided a culture of *Azot. lacticogenes*. On preliminary examination it was concluded that this species should be classified as a *Beijerinckia* and Dr. Kauffmann now agrees with this view.

2. *Azotococcus*, gen. nov., Tchan, 1953.

Cells oval, motile. Cyst is not formed; usually no acid is formed in media containing sugars.

Species typica, *Azotococcus agilis* (Beijerinck) Tchan, comb. nov., syn. *Azotobacter agilis*.

Distinguishing Characters of Different Species.

	Morphology.	Motility.	Pigmentation.	Physiology.	Habitat.
Azotococcus agilis ..	Oval. 3·5–2×2·5–2 μ	+	Green fluorescent.	Does not use mannitol or benzoate but 1% benzoate does not inhibit growth.	Water.
Azotococcus agilis var. *atypica*.	,,	,,	None.	Mannitol is a poor nutrient.	Water.
Azotococcus insigne ..	Rod 3·8–2·5× 1·6–2 μ	Cilia directly visible under dark - field microscope.	Greyish blue turn to violet.	Does not use glucose or mannitol or benzoate.	Water.

Azotococcus insigne (Derx) Tchan, comb. nov. (syn. *Azotobacter insigne*) is provisionally classified here since Derx did not mention the formation of cysts, and the aqueous habitat of the species is similar to that of *Azotococcus*.

3. *Beijerinckia* Derx, 1950.

Rods straight or slightly curved or irregular, locally swollen, characterized by the presence of highly refractive spherical bodies, presumably consisting of lipoids. No endospore. Motile or non-motile. Gram-negative. Aerobic. Nitrogen is fixed in media free of combined nitrogen. Acid is produced in media containing sugar.

Three species and one variety.

Species typica, *Beij. indica* (Starkey and De) Derx (syn. *Azotobacter indicum*).

Distinguishing Characters of Different Species.

	Pigmentation.	Motility.	Leaven Formation.	Slime Formation.	Fatty Bodies.
Beij. indica	Fulvous.	+	++	++	++
,, ,, var. *alba*	None.	±	++	++	++
,, *motile*	Amber brown.	++	−	+	Ascoccus form.
,, *lacticogenes*		−	?	−	++

Key to the genera of Azotobacteriaceae.

1. Cysts formed ..... *Azotobacter.*
2. Cysts not formed—
   (*a*) Rod with fatty bodies at each end of the cell ..... *Beijerinckia.*
   (*b*) Yeast-like oval cell ..... *Azotococcus.*

Conclusion.

The confusion in the taxonomy of *Azotobacter* is created by the contradiction between morphology, serology, physiology and chemical composition of the different species. The classification in Bergey's Manual is not acceptable. The different tests used are not satisfactory if they are used without precautions. The proposed classification is based, as for the species of *Cytophaga* (Tchan *et al.*, 1948), on the combined morphological, physiological and ecological characters. It has the advantage of leaving the